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Addendum to Quarterly Report No. 4 and Final Report Dated September 1973

THE FATE OF NITRIC OXIDE IN THE MAMMALIAN SYSTEM USING N^{15} AS TRACER AND ISOTOPIC DILUENT

Contract No. ARB 2-291

Prepared for:

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This report is an addendum to Quarterly Report No. 4 and Final Report, dated September 1973. After submitting that report, SRI was awarded an extension in time without additional funds. This addendum covers the period of the extension from September 16, 1973, to November 30, 1973.

In Quarterly Report No. 2, dated March 15, 1973, we reported that the field ionization mass spectrometer was capable of detecting $^{14}NO/^{15}NO$ ratios with a sensitivity of $\pm$ 0.01%. Thus, the identifiable quantity or concentration would correspond to about 10 $\pm$ 1 nmol of ^{14}NO/ml of whole blood.

In Quarterly Report No. 3 (June 15, 1973), we reported finding a range of 17 to 24 nmol of NO/ml of whole blood among six different individuals. There was no correlation with the smoking habits of the individual donors.

It did not seem likely that the mass ratios of 30/31 lacked specificity for NO. Alternatively, the presence of an endogenous source for the NO-hemoglobin complex was considered as a possible basis for the relative uniformity in concentrations in the human subjects.

During the period of extension of the original contract, we were able to analyze in replicate two additional human blood samples from different individuals as well as samples from two monkeys (M. speciosa) and several rats. Table 1 presents the results of these analyses.

The relatively large number of rat blood samples proved to give reproducible values and served as "negative controls" for methodology and instrumentation because they gave consistently negative results. In contrast, whereas replicate blood samples from one human donor gave repeatedly negative values (below detection limit), samples from another donor gave consistently positive results (clearly detectable) for the presence of the NO-hemoglobin complex. Neither donor had any unusual exposure to environmental NO. The two monkey blood samples also showed the presence of NO-hemoglobin.

Therefore, a very important question remains: To what metabolic, genetic, or environmental factor(s) in the donors does the presence or absence of the NO-hemoglobin complex in human blood relate? It appears that there are exceptions among people to the relative uniformity of blood levels indicated earlier.

In SRI Proposal LSH 73-123 (August 15, 1973) submitted to the Air Resources Board of the State of California, we suggested investigations that could lead to the understanding of the nature and significance of the NO-bound hemoglobin.

Table 1

FIELD IONIZATION MASS SPECTROMETRIC ANALYSIS
OF BLOOD SAMPLES FOR NO-HEMOGLOBIN COMPLEX

Sample	Mass 30/31 Ratio* (percent)	Nanomoles of ^{14}NO per Milliliter of Blood	Mean	SD ±	SE ±	"p"
Human						
GF - 1	0.263	14.8				
2	0.236	0.1				
3	0.231	7.2	5.8	5.2	2.1	
4	0.221	1.9				
5	0.226	4.6				
6	0.229	6.6				<0.001
LJ - 1	0.280	24.1				
2	0.291	30.2	26.9	3.1	1.7	
3	0.260	26.3				
Primate						
A	0.271	19.3	22.3	4.2	3.0	
B	0.282	25.3				
Rodent						
A - 1	0.252	8.8				<0.002
2	0.232	0.1				
3	0.241	2.7				
B - 1	0.236	0.1				
2	0.259	12.6				
C - 1	0.231	7.67	5.7	4.8	1.5	
2	0.235	9.63				
D - 1	0.242	10.9				
2	0.226	4.6				
E	0.217	0.1				

*The mean of seven blank (air only, without blood; see Quarterly Report No. 2) values for the mass 30/31 ratios was 0.230 ± 0.014.